Effect of Heat Processing and Storage Time on Migration of Bisphenol A (BPA) and Bisphenol A−Diglycidyl Ether (BADGE) to Aqueous Food Simulant from Mexican Can Coatings

Elvia M. Munguia-Lopez and Herlinda Soto-Valdez*

Centro de Investigacion en Alimentacion y Desarrollo, A.C. Apartado Postal 1735, Hermosillo, Sonora 83000, Mexico

Effects of heat processing and storage time (up to 70 days) on migration of bisphenol A (BPA) and bisphenol A−diglycidyl ether (BADGE) from can coatings into an aqueous food simulant were determined. Distilled water was canned in two types of Mexican cans: for tuna and for jalapeño peppers. Results showed that there is an effect of heat treatment on migration of both compounds. Storage time did not show any effect in BPA migration from tuna cans. There was an effect of storage time on BPA migration from jalapeño pepper cans. Results for BADGE migration were affected by its susceptibility to hydrolyze in aqueous simulants. BADGE concentration decreased, or was not detected, during storage in both types of cans. Migration levels for BPA and BADGE were within 0.6−83.4 and <0.25−4.3 μg/kg, respectively. Both were below European and Mercosur legislation limits. Other migrating compounds were detected, although no identification was performed.

Keywords: *Bisphenol A; bisphenol A−diglycidyl ether; BADGE; epoxy resins; canned food*

INTRODUCTION

Metallic cans are protected against corrosion by the application of inner coatings based on epoxy or organosol type resins (*1*). If coatings are inadequately formulated, they can be a source of contamination due to the migration of chemicals into the food (*2*). The synthesis of bisphenol A (BPA) type epoxy resins is shown in Figure 1. Condensation between BPA and epichlorhydrin yields bisphenol A−diglycidyl ether (BADGE) and polymers of different molecular masses. These epoxy resins are soluble and fusible. It is necessary to add curing agents and high temperature to obtain a thermosetting product that is stable enough to protect metallic cans (*3*). BPA or BADGE may remain unreacted when polymerization conditions or the curing process is insufficient. Organosol resins are manufactured by mixing several types of resins. They may contain poly(vinyl chloride) (PVC), epoxy, phenolic, and epoxyphenolic resins. PVC undergoes degradation at curing temperatures giving rise hydrochloric acid (HCl). Organosols are stabilized with epoxy type compounds such as BADGE, which is a scavenger for HCl (Figure 2) (*4*). Residual BPA and BADGE in coatings are prone to transfer to canned food.

The toxicity of BPA is related to estrogenic activity. It is among estrogenic xenobiotics that may affect the reproductive system of animals and causes proliferation of breast cancer cells in vitro (*5*, *6*). The toxicity of BADGE is related to cytotoxic effects in tissues with a high rate of cell division. It is listed as a tumorigen, mutagen, and primary irritant by the U.S. National Institute for Occupational Safety and Health (*6*). The European Commission is considering extension of the current legislation on plastics for food contact to surface coatings on cans. At the moment, migration limits for BPA and BADGE are 3 mg/kg (*7*) and 1 mg/kg (*6*) of food or food simulant, respectively. Mercosur legislation on food contact materials set a migration limit for BPA (3 mg/kg of food or food simulant) but not for BADGE (*8*).

Brotons et al. (*9*) reported estrogenic activity (determined by E-screen bioassay) from canned peas, artichokes, and mixed vegetables. They found that BPA was transferred to water after a second heat process was applied to cans in which the original contents had been removed. BADGE has been reported to migrate to canned fatty foodstuffs in European countries. Levels of migration were found to be mainly below the European limit. However, up to 16% of fish samples have been found to exceed such limits, mainly in sardines and anchovies (*10*−*12*). Migration of BADGE to aqueous food simulants has not been found to exceed the maximum permitted levels (*3*, *13*). Because it shows instability in aqueous media, information about hydrolysis products and kinetics of decomposition of BADGE has been published (*14*−*16*). None of the above references reported migration during long storage time periods.

Tuna and jalapeño peppers are among the most consumed canned foodstuffs in Mexico. Tuna in brine demand is increasing because of health reasons. Presently, there is no legislation for food contact materials in Mexico. There are no reports about the quality of resins used in the Mexican canning industry from the migration point of view. The aim of this work was to determine the effect of heat processing and storage time on the migration of BPA and BADGE in an aqueous food simulant from two types of cans frequently used in Mexico.

* Author to whom correspondence should be addressed (fax 52 62 80 0422; telephone 52 62 89 2400; e-mail hsoto@cascabel.ciad.mx).

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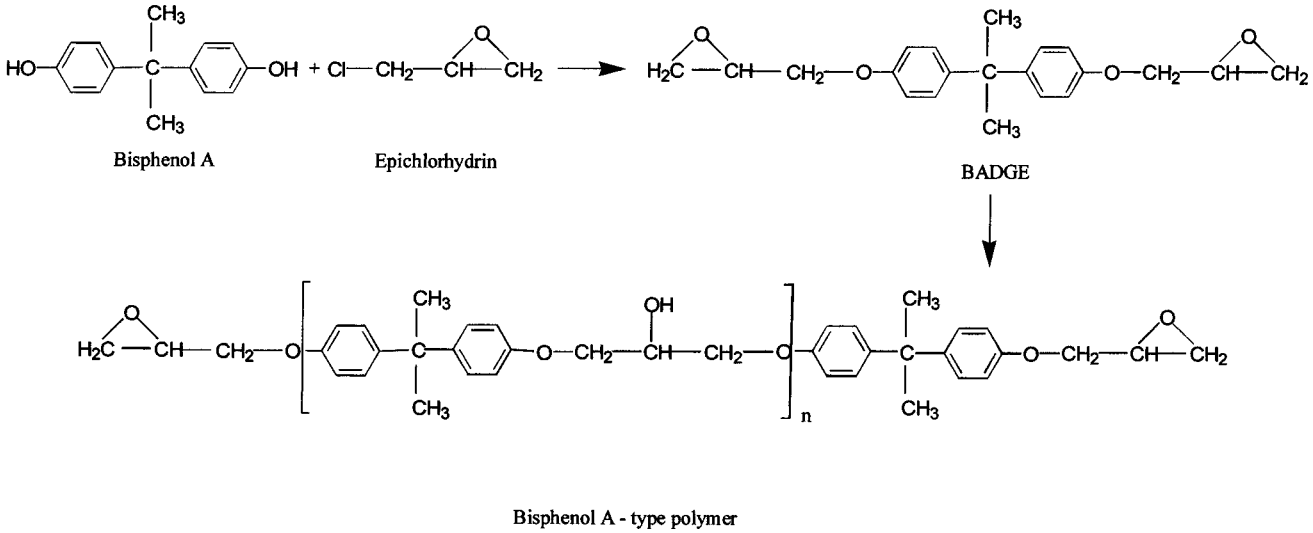

Figure 1. Synthesis of bisphenol A type resins.

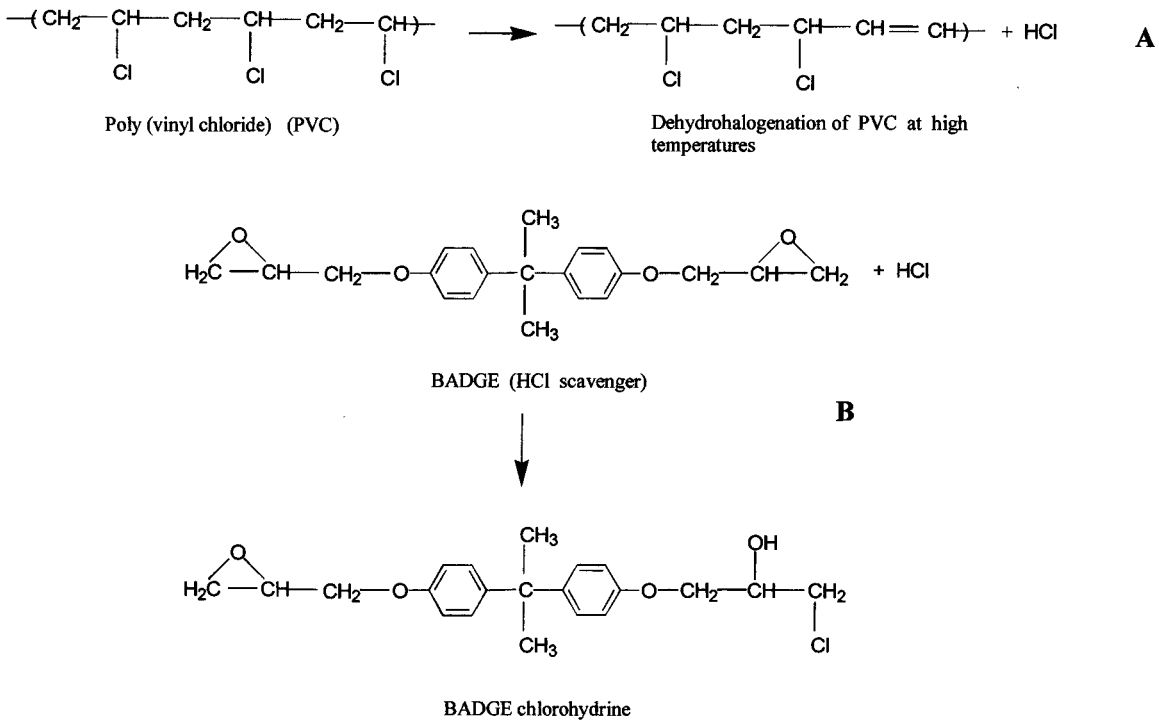

Figure 2. Degradation of PVC at curing temperatures (A); BADGE reaction with HCl (B).

MATERIALS AND METHODS

Chemical and Reagents. BPA standard was of analytical grade (Aldrich, Milwaukee, WI). BADGE was provided by Shell Chemical Co., Houston, TX. Its purity was confirmed to be 99% by using a BADGE standard provided by a member of European groups working in food contact materials (Departamento de Quimica Analitica, Centro Politecnico Superior de Ingenieros, Universidad de Zaragoza, Zaragoza, Spain). Distilled water was used as the aqueous food simulant. Acetonitrile (ACN) was of HPLC grade (EM Science, Gibbstown, NJ). Water for chromatographic analyses was of HPLC grade (J. T. Baker, Xalostoc, Mexico).

Apparatus. An infrared spectrophotometer FTIR Nicolet, Protege 460 (Nicolet, Madison, WI) was used to identify the can coatings. Scanning conditions were as follows: wavenumber range, from 4000 to 400 cm^{-1}; resolution, 4 cm^{-1}; number of scans, 64; scan speed, 0.63; detector, DTGS. An Aldrich Library of Infrared Spectra was used for identification (*17*). A high-pressure liquid chromatographic system (HPLC) was used for quantification of BPA and BADGE (Varian, Star 9012, pump, Mexico D.F., Mexico) equipped with a fluorescence detector (Varian 9075) and a Star 5 chromatography workstation (Varian). BPA and BADGE identities were confirmed by gas chromatography−mass spectrometry (GC-MS) using a Varian 3400CX gas chromatograph coupled to a Saturn III model mass spectrometer (Varian), with an NIST 92 data system. *Spectra for the Identification of Monomers in Food Packaging* was used for identification (*18*).

Liquid Chromatographic Conditions. Fluorescence detector excitation wavelength was 224 nm, and emission wavelength was 310 nm. A 15 cm × 5 mm i.d. Micropak C_{18} MCH-5-N-CAP column (Varian), protected by C_{18} guard columns, was used. A Rheodyne 7125 injector with a 10 μL loop was used. A 1 mL/min flow with the following gradient program was used: 10 min isocratic elution with ACN/water (35:65), 5 min linear gradient to 50:50, 5 min isocratic elution,

10 min gradient elution to 60:40, 5 min isocratic elution, 5 min gradient elution to 100:0, and 5 min isocratic elution. Calibration curves for BPA and BADGE were prepared from 25 to 1200 μg/L in ACN. Under these chromatographic conditions, retention times for BPA and BADGE were between 16.8 and 17.1 and between 31.5 and 32.0 min, respectively.

Gas Chromatographic Conditions. A gas chromatograph was fitted with a DB5 fused silica capillary column (30 m × 0.25 mm i.d., film thickness = 0.25 μm) supplied by J&W Scientific (Supelco, Mexico, D.F., Mexico). The oven temperature was programmed from 90 to 300 °C at 30 °C/min and held for 10 min. The injector temperature was 150 °C, and that of the transference line was 200 °C. Under these chromatographic conditions, retention times for BPA and BADGE were 8.5 and 11.6 min, respectively.

Migration Tests. Two types of cans, each from the same batch, were tested. Tuna cans (18 items) were provided by Productos Pesqueros de Guaymas, S.A. de C.V., and jalapeño pepper cans (18 items) by Herdez, S.A. de C.V. Internal coatings were identified by infrared spectrophotometry. Distilled water, as the aqueous food simulant, was canned in both types of cans. Heat processing (HP) was applied to half of each batch (9 items). Temperature and time was similar to those applied during the processing of tuna (121 °C/90 min) and jalapeño peppers (100 °C/9 min). No heat process (NHP) was applied to the remaining cans (9 items). All cans were stored at 25 °C, and samples (three cans) were taken at 0, 40, and 70 days. NHP cans for 0 days of storage were in contact with resin for ~4 h before analysis.

BPA and BADGE Analyses: Each can sampled was opened and water evaporated by rotary evaporator at temperature up to 35 °C. Dry residue was redissolved in 5 mL of ACN and filtered through 0.22 μm Durapore membrane filters (Millipore Cork, Ireland) before injection into the HPLC system.

RESULTS AND DISCUSSION

Inner coatings were identified, by FTIR, as organosol resin (Figure 3A) and epoxy resin (Figure 3B) in tuna and pepper cans, respectively (*17*). Characteristic absorption bands for organosol resin occur at about 1427, 703, and 612 cm^{-1}. The epoxy resin spectrum shows typical aromatic bands at 3040, 1605, 1508, and 826 cm^{-1}. Both resins contain an ester type additive, identified by bands at 1722−1730 and 1236−1244 cm^{-1}. These results agree with those of Summerfield et al. (*10*) for different brands of anchovy cans.

HPLC chromatogram A in Figure 4 was obtained from a standard solution of both BPA and BADGE. HPLC chromatogram B corresponds to an aqueous simulant sample from tuna cans in ACN. HPLC chromatogram C corresponds to an aqueous simulant sample from pepper cans in ACN.

Recoveries were 100.1 ± 4.7 and 93.58 ± 4.38% for BPA and BADGE, respectively. They were calculated by spiking the aqueous simulant from a can to get a 300 μg/L solution of both compounds. Regression coefficients for calibration lines were 0.983 and 0.998, and instrument detection limits were 20 and 25 μg/L for BPA and BADGE, respectively. The procedure used in this work was able to quantify migration levels as low as 0.20 and 0.25 μg/L for BPA and BADGE, respectively.

Effect of Heat Processing and Storage Time on Migration of BPA. Table 1 shows the concentration of BPA found in both types of cans during storage. As was expected, there was an effect of heat processing on migration. A drastic heat process (121 °C/90 min) is applied to tuna for commercial sterilization because it is a low-acid food. Migration of BPA remained constant during 0, 40, and 70 days of storage for HP tuna cans. Therefore, there was no effect of storage time in BPA

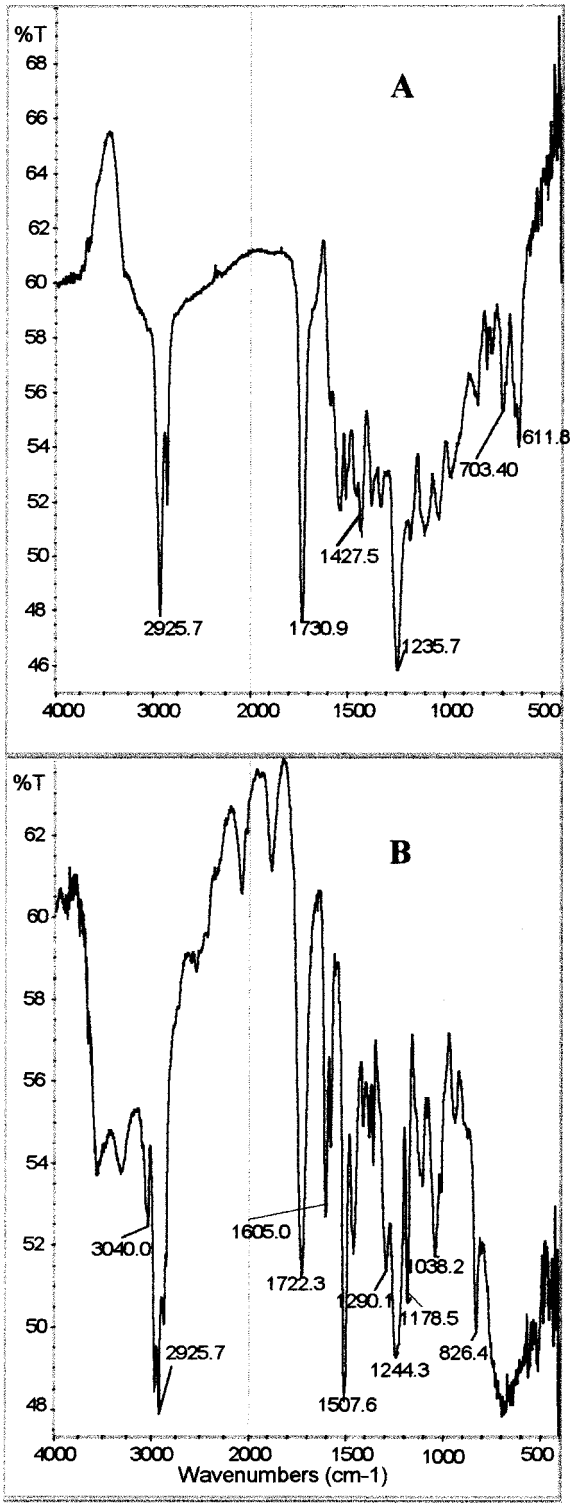

Figure 3. IR spectra of inner coatings used in tuna (A) and jalapeño pepper (B) cans.

migration. No references for BPA content in organosols have been reported. They may contain BADGE and epoxy resins (*4*). It may come from a BPA and BADGE mixture or a rich BPA epoxy resin, which is added to the organosol resin used for tuna cans. However, it seems these additives are not very high in proportion in this organosol because the IR spectrum (Figure 3A) did not show any aromatic band at >3000 cm^{-1}. In this work, BPA was confirmed in selected samples by GC-MS. MS spectra showed main ion fragments at *m/z* 213, 228, 119, and 91 (*18*).

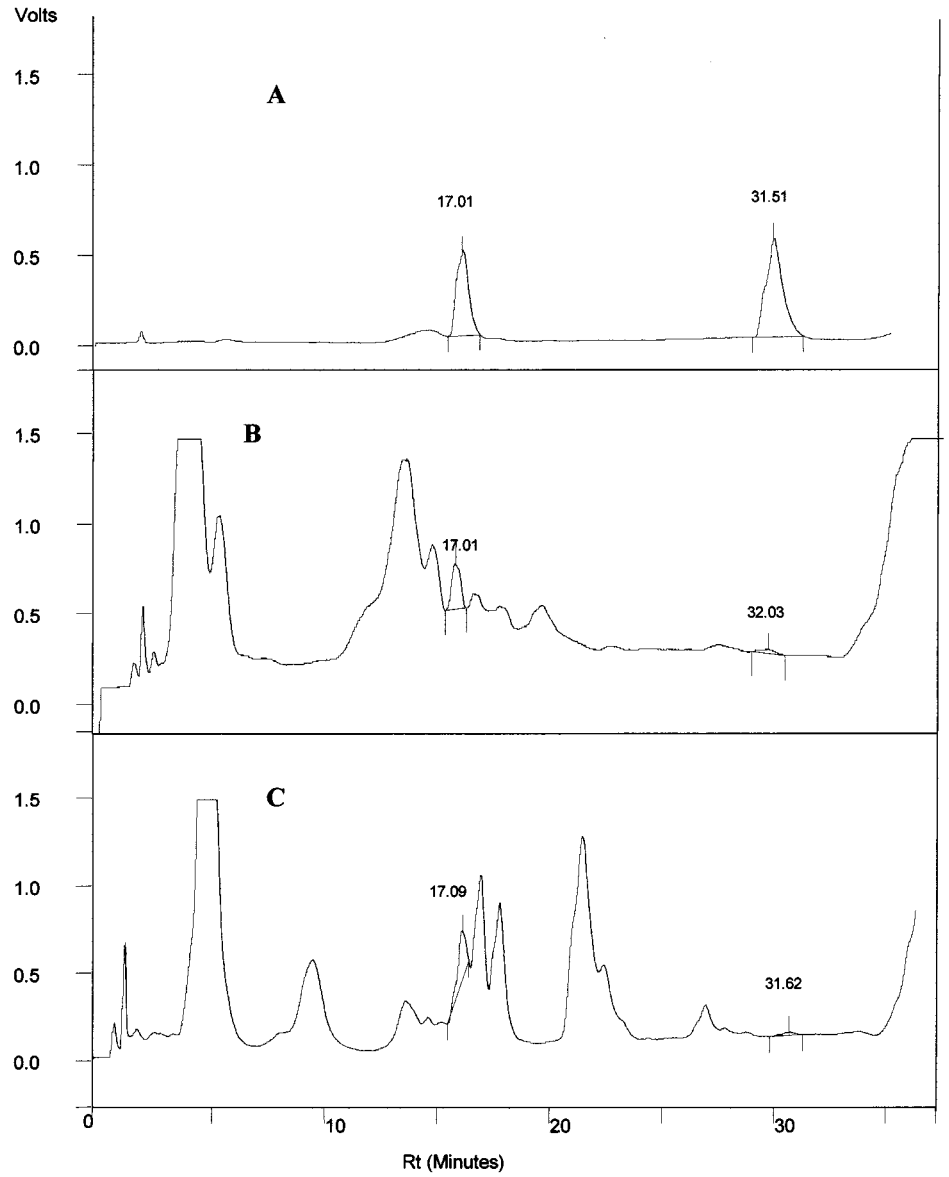

Figure 4. HPLC chromatograms: (A) standard solution of 1200 μg/L of BPA and BADGE; (B) aqueous simulant from tuna cans, redissolved in ACN; (C) aqueous simulant from jalapeño pepper cans, redissolved in ACN.

Table 1. Migration of BPA from Tuna and Jalapeño Pepper Cans to Water as Aqueous Food Simulant

	μg/kg BPA			
storage time (days at 25 °C)	tuna cans[a]		jalapeño pepper cans[b]	
	NHT[c]	HT[d]	NHT[c]	HT[e]
0	<0.20	81.80 ± 40.00	<0.20	1.5 ± 0.38
40	14.5 ± 0.90	83.40 ± 18.00	0.58 ± 0.11	3.42 ± 0.54
70	12.5 ± 1.30	79.70 ± 11.00	4.20 ± 0.54	4.16 ± 0.54

[a] Organosol resin coated. [b] Epoxy resin coated. [c] No heat process applied. [d] Heat process applied, 121 °C/90 min. [e] Heat process applied, 100 °C/9 min.

Heat processing conditions are milder for peppers (100 °C/9 min) than for tuna because of their low pH. In this case, part of the residual BPA remained on the coating after processing. Consequently, BPA concentration increased during storage time (Table 1). NHP cans showed no migration at zero time; however, it was detected at 40 and 70 days. Tuna cans showed higher migration of BPA than peppers cans, except for the 70 days of storage. Migration levels for BADGE in organosols have been reported to be higher than those in epoxy resins, but no reports have been found for BPA. Levels of up to 83.4 μg/kg of BPA were found in aqueous simulant, which are far lower than European and Mercosur limits of migration. It was out of the scope of this work to determine estrogenic activity. However, these levels can be assumed to show estrogenic activity by comparison to levels of BPA found in canned peas, artichokes, and mixed vegetables (22−76 μg/kg) by Brotons et al. (*9*). They found that 76 μg/kg of BPA in a pea extract showed 58% estrogenic effect compared to that of a 10 pM solution of estradiol 17-β.

Effect of Heat Processing and Storage Time on Migration of BADGE. Table 2 shows the concentration of BADGE found in both types of cans during storage. The BADGE concentration decreased during storage time in both types of HP cans. It disappeared at 40 days of storage for HP tuna cans. BADGE decreased with time at levels from 2.7 to 0.29 μg/kg in HP pepper cans. Most of the NHP cans showed BADGE migration below the limits of the procedure used. The above behaviors may not depend on heat processing or storage time but on BADGE stability. BADGE hydrolyzes in aqueous

Table 2. Migration of BADGE from Tuna and Jalapeño Pepper Cans to Water as Aqueous Food Simulant

storage time (days at 25 °C)	μg/kg BADGE			
	tuna cans[a]		jalapeño pepper cans[b]	
	NHT[c]	HT[d]	NHT[c]	HT[e]
0	<0.25	4.30 ± 0.73	<0.25	2.70 ± 0.47
40	<0.25	<0.25	0.37 ± 0.13	0.76 ± 0.17
70	<0.25	<0.25	<0.25	0.29 ± 0.10

[a] Organosol resin coated. [b] Epoxy resin coated. [c] No heat process applied. [d] Heat process applied, 121 °C/90 min. [e] Heat process applied, 100 °C/9 min.

medium (*16*). Hydrolysis products were not determined because when the experiment was carried out, there were no commercial standards available. Information about hydrolysis products and kinetics of decomposition of BADGE has been published (*14*−*16*). None of these works found levels of BADGE exceeding the maximum permitted levels in any aqueous food simulants. However, higher levels of BADGE migration have been found in fatty foodstuffs. Summerfield et al. (*10*) reported BADGE levels in 181 retail samples of European canned foodstuffs. BADGE exceeded the limit tolerated by European legislation (1 mg/kg) in 5 of the 22 sardine samples analyzed and 7 of the 15 anchovy samples. In Austria 67 canned foodstuffs were analyzed for BADGE (*11*); 16% of all samples were found to be above the limit. Simoneau et al. (*12*) determined BADGE in 382 fish samples, finding 3% exceeding the limit. In this work, BADGE was confirmed in selected samples by GC-MS. MS spectra showed main ion fragments at *m*/*z* 340, 325, 269 and 213 (*18*).

Other Migrating Compounds. Chromatograms B and C in Figure 4 correspond to aqueous simulant samples from tuna and pepper cans. Several signals corresponding to different migrants were found. Under the analytical conditions of this work, some of these signals were of higher intensity than BPA and BADGE. Identification of these signals was out of the scope of this work. Hydrolysis products of BADGE may appear among these signals because they have been identified as BADGE mono-ol epoxide and BADGE diol epoxide in water (*2*, *19*). Other authors have reported to have identified a cyclic dimer of BADGE in several samples of canned fish products; linear dimer and trimer were also identified in one sample (*20*). Epoxy resin components such as curing agents, lubricants, and other additives may be among the signals detected. Due to the presence of other migrants from can coatings, Grob et al. (*21*) called for more effective regulations of coatings with food. Identification and toxicological tests have to be performed with the unidentified migrants to ensure the quality of can coatings. Legislation for food contact materials should be implemented in Mexico, including those compounds unidentified in this work.

Conclusions. Migration levels of BPA and BADGE from two types of Mexican tuna and jalapeño pepper can coatings to aqueous simulant did not exceed the European and Mercosur legal limits.

BPA migration from organosol resin in tuna cans was higher than that from epoxy resin in jalapeño pepper cans.

Heat processing enhanced the rate of migration for both compounds in both types of cans. Storage time did not show any effect in BPA migration from tuna cans. There was an effect of storage time on BPA migration from jalapeño pepper cans. BADGE migration was affected by its susceptibility to hydrolyze in aqueous simulants.

There are other migrating compounds that could be in higher concentrations than BPA or BADGE in aqueous food simulants in this work. They should be considered in legislation related to food contact materials.

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Supporting Information Available: HPLC chromatogram and MS spectra. This material is available free of charge via the Internet at http://pubs.acs.org.

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